UNITED STATES PATENT AND TRADEMARK OFFICE

BEFORE THE BOARD OF PATENT APPEALS AND INTERFERENCES

Ex parte LARRY A. SKLAR, ERIC R. PROSSNITZ, JANEEN VILVEN and DONNA NELDON

Appeal No. 2004-1728
Application No. 09/370,358

ON BRIEF

Before SCHEINER, MILLS and GRIMES, Administrative Patent Judges.

SCHEINER, Administrative Patent Judge.

DECISION ON APPEAL

This is a decision on appeal under 35 U.S.C. § 134 from the examiner's final rejection of claims 1-6, 8-13, 15-17, 48, 51 and 53-57; claims 14 and 18-47 are also pending but have been withdrawn from consideration.

Claims 1, 2, 48 and 54 are representative of the subject matter on appeal:

1. A method for non-cellular analysis and display of 7-transmembrane receptors comprising the following steps:
a) incorporating an attachment tether to a receptor;
b) solubilizing the receptor; and
c) presenting the receptor in conjunction with a support, wherein said support comprises at least one substrate selected from the group consisting of silica bead substrates, latex bead substrates and other bead substrates appropriate for flow cytometry, and wherein the receptor in conjunction with a support is analyzed with a flow cytometer in real-time.

2. The method of claim 1 wherein the step of incorporating an attachment tether to a receptor comprises incorporating at least one of the following tags from the group consisting of C-Histidine, N-Histidine, biotin, and GST tags.

48. A method for non-cellular display of 7-transmembrane receptors comprising the following steps:

a) incorporating an attachment means to a receptor;

b) solubilizing the receptor;

c) presenting the receptor in conjunction with a support;

d) presenting at least one ligand to bind to the receptor, wherein said ligand is known to bind to the receptor;

e) combining the receptor and ligand to accomplish binding while the receptor is bound to the support; and

f) sorting the bound receptor ligand pairs by fluorescence and using flow cytometry to analyze the fluorescence and binding interactions in real-time.

54. The method of claim 1 wherein the step of incorporating an attachment tether to a receptor comprises incorporating at least one epitope tag.

The references relied on by the examiner are:

Baumbach et al. (Baumbach)	5,583,010	Dec. 10, 1996
Dower et al. (Dower)	5,639,603	Jun. 17, 1997

Robeva et al. (Robeva), "Double Tagging Recombinant A_1 - and A_{2A} -Adenosine Receptors with Hexahistidine and the FLAG Epitope," Biochemical Pharmacology, Vol. 51, pp. 545-555 (1996)

Claims 1, 6, 9-13, 15-17, 48, 51 and 53-57 stand rejected under 35 U.S.C. § 103 as unpatentable over Baumbach and Dower. Claims 2-5 and 8 stand rejected under 35 U.S.C. § 103 as unpatentable over Baumbach, Dower and Robeva.

We reverse.

BACKGROUND

> One of the largest families of receptors in the human genome is that of the 7-transmembrane [receptor] (7-TMR) superfamily, also known as G-protein [(guanine nucleotide-binding protein)] coupled receptors . . .
>
> 7-TMR have seven transmembrane α-helical domains [ ], with three connecting loops on each inner and outer face of membrane . . . The three extracellular loops and transmembrane region participate in ligand binding . . . The intracellular loops . . . and tail, in contrast, participate in

> interactions with the G-protein . . . The pathway of cell activation for monovalent chemoattractant ligands appears to involve the interaction of receptor-ligand complexes with [G-proteins].

Specification, page 2.

According to appellants, "traditional methods for examining receptor behavior require a separation step, frequently involving centrifugation or filtration" but "[t]hese steps are not optimal for real-time kinetic analysis of rapidly equilibrating systems" (id.). The present invention, on the other hand, is directed to "non-cellular display of 7-transmembrane receptors comprising the steps of incorporating an attachment scheme to a receptor, solubilizing the receptor, and presenting the receptor in conjunction with a support . . . [p]referably . . . by affinity coupling the receptor to a . . . bead substrate appropriate for flow cytometry" (id., page 4).

The specification teaches that attachment schemes include incorporation of a C-histidine, N-histidine, biotin, or GST tag into an oligonucleotide [receptor] construct prior to expression and solubilization of the receptor (Specification, page 4), or addition of an epitope tag to the receptor (id., Example 5 and Figure 6). The specification also teaches that receptors "bound to beads using either the N-His or C-His tags, [ ] are able to bind fluorescent ligand" (id., page 9), and "[t]he N-His receptor can be used where the 'extracellular face' of the receptor is in proximity to the bead and the intracellular face is away from the bead and consequently available to the G-protein" (id.).

According to appellants, "the display and assays provided by the present invention allow an important sequence of signaling events (ligand binding, receptor and G-protein coupling, and receptor desensitization) to be evaluated" in real-time (Specification, page 4).

DISCUSSION

The examiner rejected claims 1, 6, 9-13, 15-17, 48, 51 and 53-57 under 35 U.S.C. § 103 as unpatentable over Baumbach and Dower. Baumbach describes the isolation, characterization and cloning of a growth hormone releasing hormone receptor (GHRH-R), a member of the 7-transmembrane receptor family. Briefly, solubilized GHRH-R-GHRH (receptor-ligand) complexes are dissociated to obtain a partially purified GHRH-R isolate, "capable of being used to determine the amino acid residue sequence of the GHRH-R" and to "provide[ ] for the cloning of a gene for a GHRH-R" (column 7, line 48 to column 8, line 17). Baumbach broadly describes measuring receptor-ligand binding in "[s]creens us[ing] the purified or recombinant receptor in solid or liquid phase" (column 19, lines 65-67).

> Expression in the screen[s] is measured by ligand binding, second messenger function, product secretion, or by other means. Solid phase assays can involve receptor attached to a solid support either chemically or immunologically in conjunction with or without transduction proteins. These assays can be linked to a reporter such as antibody, biological chemical (e.g., biotin), or binding protein or enzyme, which will express a radioactive, chemical, calorimetric or luminescent signal.

Column 19, line 67 to column 20, line 7.

The examiner concedes that Baumbach is "silent with regard to flow cytometry," but argues that Dower "teaches the general applicability of flow cytometry to the sorting of isolated solubilized receptors and their bound ligands, wherein the solid supports are beads appropriate for library screening" (Answer, page 4). The examiner points to Dower's basic flow cytometric assays for screening combinatorial libraries for molecules able to bind a receptor of interest. One assay (Dower, column 31, lines 57-63) involves

> binding of a receptor labelled with a fluorescent tag to a mixture of beads displaying the diverse molecules of a molecular library. After washing away unbound or non-specifically bound receptors, one then employs [flow cytometry] to sort the beads and to identify and isolate physically individual beads showing high fluorescence.

In another assay (Dower, column 36, lines 35-42),

> [a] tagged oligomer in solution can be assayed using a receptor immobilized by attachment to a bead, for example, by a competition assay with a fluorescently labeled ligand. One may recover the beads bearing immobilized receptors and sort the beads using [flow cytometry] to identify positives (diminished fluorescence caused by the library molecule competing with the labeled ligand).

According to the examiner, "it would have been obvious to one of ordinary skill in the art . . . to use beads as the solid support [in Baumbach's assay] and to separate the receptor-ligand pairs by flow cytometry" because Dower teaches "by adopting cell sized solid supports or beads, one can use flow cytometry for high sensitivity receptor binding analysis and facile bead manipulation" (Answer, page 5).

Appellants argue that the examiner's rejection "based on Baumbach in view of Dower is prima facie improper because the stated motivation for combining Dower with Baumbach is found in Dower not Baumbach. For a proper rejection to be based on Baumbach in view of Dower, the motivation should be found in Baumbach not Dower . . . [t]herefore, Dower cannot be properly combined with Baumbach" (Brief, page 6). Along the same lines, appellants argue that "Dower 'teaches the general applicability of flow cytometry to the sorting of isolated solubilized receptors and their bound ligands'," while Baumbach "is silent with respect to flow cytometry" (id., pages 6-7). "Therefore, a person of ordinary skill in the art just possessing Dower would have no motivation to look to Baumbach" (id., page 6).

We know of no authority that would support appellants' position in this matter. As the examiner correctly points out, "[t]he test for an implicit showing is what the combined teachings, knowledge of one of ordinary skill in the art, and the nature of the problem to be solved as a whole would have suggested to those of ordinary skill in the art." In re Kotzab, 217 F.3d 1365, 1370, 55 USPQ2d 1313, 1317 (Fed. Cir. 2000). Both Baumbach and Dower concern analysis of solubilized, subsequently immobilized receptors, and their ligands. Baumbach broadly describes analysis of receptor-ligand binding using receptor immobilized on a solid phase (Baumbach, column 19, lines 65-67); Dower describes analysis of receptor-ligand binding by flow cytometry using receptor immobilized on a bead (Dower, column 36, lines 35-42). Like the examiner, we know of no reason why the combination of the teachings of these two references should be improper.

This brings us to appellants' argument that "the general methodology and the flow cytometric analysis of Dower is very different from that recited in claim 1" in that "Dower solubilizes and binds a ligand library to a bead . . . [while] claim 1 recites the solubilization and subsequent attachment via a tether of a receptor to a bead" (Brief, page 8). However, as discussed above, and as pointed out by the examiner, Dower additionally specifies that "[a] tagged oligomer is solution can be assayed using a receptor immobilized by attachment to a bead" (Dower, column 36, lines 35-37).

Nevertheless, appellants also argue that "the Baumbach and Dower [references] fail to teach or suggest analyzing with a flow cytometer in real-time as recited in [the] claim[s]" (Brief, page 8), as illustrated by Dower's requirement for "a washing step to remove unbound or non-specifically bound receptors before analyzing with a flow

cytometer" (id.). In our view, the examiner has not adequately addressed this issue. We note the examiner's assertion that "one of ordinary skill in the art appreciates that, even with the wash step (exclusion of which is not a positive claim limitation), Dower teaches analyzing with flow cytometry in real time, i.e. the disassociation of the ligand and the receptor is happening as they are being analyzed in the flow cytometer" (Answer, page 8), but we cannot agree with this interpretation of Dower's teachings. Ostensibly, Dower's wash step illustrates that the receptor-ligand binding step is separated in time from the analysis step (apparently analysis occurs sometime after equilibrium is reached). It may be that a certain amount of disassociation occurs after the wash step, but the examiner does not identify anything in Dower which would indicate that binding interactions are measured as they occur, i.e., in real-time.

Then there is the matter of the step requiring incorporation of an "attachment tether" or "attachment means" prior to solubilization and display of the receptor on a bead (see claims 1 and 48). As taught in the specification, attachment tethers include C-Histidine, N-Histidine, biotin, GST and epitope tags (see pages 4, 9 and 16). It is clear from Example 5 and corresponding Figure 6 of the specification that a "tag" is not an integral part of the receptor, but is a discrete moiety added to the receptor (see "tag" (26) of Figure 6, and also compare "receptor" (18) with "epitope tagged receptor" (20)). Dower does not appear to describe any sort of attachment tether, and we disagree with the examiner that Baumbach's mention of receptors "attached to a solid support either chemically or immunologically" meets the requirement for a discrete attachment tether.

"[T]the examiner bears the initial burden of presenting a prima facie case of obviousness." In re Rijckaert, 9 F.3d 1531, 1532, 28 USPQ2d 1955, 1956 (Fed. Cir. 1993). "It is impermissible to use the claimed invention as an instruction manual or 'template' to piece together the teachings of the prior art so that the claimed invention is rendered obvious." In re Fritch, 972 F.2d 1260, 1266, 23 USPQ2d 1780, 1784 (Fed. Cir. 1992). The fact that the prior art could have been modified in a manner consistent with appellant's claims would not have made the modification obvious unless the prior art suggested the desirability of the modification. In re Gordon, 733 F.2d 900, 902, 221 USPQ 1125, 1127 (Fed. Cir. 1984).

On this record, the only reason or suggestion to display the receptor on a bead using an attachment tether, or to analyze receptor interactions in real-time, comes from appellants' specification, rather than the combined teachings of Baumbach and Dowry. Accordingly, we are constrained to reverse the rejection of claims 1, 6, 9-13, 15-17, 48, 51 and 53-57 under 35 U.S.C. § 103.

Claims 2-5 and 8 stand rejected under 35 U.S.C. § 103 as unpatentable over Baumbach, Dower and Robeva. Robeva describes the introduction of FLAG-epitope and hexahistidine tags into recombinant adenosine receptors to facilitate purification of the receptors using anti-FLAG and Ni-NTA affinity columns. According to the reference, addition of the tags "has little effect on ligand binding and appears not to disrupt G protein coupling" (Robeva, page 554, left-hand column).

According to the examiner, it would have been obvious "to use histidine tags . . . as the attachment means for the assay disclosed by [Baumbach] modified according to

the teachings of [Dower]" because Robeva "state[s] that [the] method should be useful for . . . a variety of methods including reconstitution assays" (Answer, page 6).

As with the previous rejection, however, the only reason or suggestion to analyze receptor interactions in real-time comes from appellants' specification, rather than the combined teachings of Baumbach, Dowry and Robeva. Accordingly, the rejection of claims 2-5 and 8 under 35 U.S.C. § 103 is reversed as well.

REVERSED

Toni R. Scheiner
Administrative Patent Judge

Demetra J. Mills
Administrative Patent Judge

Eric Grimes
Administrative Patent Judge

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